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## Review

# Selection and breeding of plant cultivars to minimize cadmium accumulation

C.A. Grant<sup>a,\*</sup>, J.M. Clarke<sup>b</sup>, S. Duguid<sup>c</sup>, R.L. Chaney<sup>d</sup>

<sup>a</sup>AAFC Brandon Research Centre, Box 1000A, R.R.#3, Brandon, MB Canada, R7A 5Y3

<sup>b</sup>AAFC Semiarid Prairie Agricultural Research Centre, Swift Current, SK, Canada S9H 3X2

<sup>c</sup>AAFC Morden Research Station, Morden, MB, Canada R6M 1Y5

<sup>d</sup>USDA, ARS, Animal Manure and Byproducts Laboratory, Room 013, Building 007, BARC-West, 10300 Baltimore Avenue, Beltsville, MD 20705-2350, USA

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## ABSTRACT

Natural variation occurs in the uptake and distribution of essential and nonessential trace elements among crop species and among cultivars within species. Such variation can be responsible for trace element deficiencies and toxicities, which in turn can affect the quality of food. Plant breeding can be an important tool to both increase the concentration of desirable trace elements and reduce that of potentially harmful trace elements such as cadmium (Cd). Selection programs for a low-Cd content of various crops, including durum wheat, sunflower, rice and soybean have been established and low-Cd durum wheat cultivars and sunflower hybrids have been developed. In durum wheat (*Triticum turgidum* L. var *durum*), low-Cd concentration is controlled by a single dominant gene. The trait is highly heritable, and incorporation of the low-Cd allele can help to reduce the average grain Cd to levels below proposed international limits. The allele for low-Cd concentration does not appear to affect major economic traits and should not cause problems when incorporated into durum cultivars. The cost of Cd selection in a breeding program is initially large both in terms of Cd determination and reduced progress towards development of other economic traits, but declines as more breeding lines in the program carry the low-Cd trait and are utilized in new crosses. Production of low-Cd crop cultivars can be used as a tool to reduce the risk of movement of Cd into the human diet.

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\* Corresponding author. Tel.: +1 204 578 3570; fax: +1 204 728 3858.

E-mail address: [cgrant@agr.gc.ca](mailto:cgrant@agr.gc.ca) (C.A. Grant).

## 1. Introduction

The beneficial and adverse effects of trace element content in the diet on human health are particularly great in staple food crops that make up a large proportion of dietary intake. Natural variation occurs in both the uptake and the distribution of essential and nonessential trace elements in crop species and in cultivars within species (Clarke et al., 1997a; Bell et al., 1997; Graham and Welch, 1999; Huang and Graham, 2001; Graham et al., 2007). The accumulation and distribution of trace elements in plants is affected by factors under genetic control. These factors include size and morphology of the root system, production of root exudates that can mobilize elements, root association with microorganisms such as mycorrhizal fungi and root-colonizing bacteria, the amount and activity of transport systems across cell membranes, presence of intracellular binding sites, vacuolar sequestration, xylem loading and unloading, phloem transport and retention in the root (Harris and Taylor, 2004; Ishikawa et al., 2005a; Arao and Ishikawa, 2006). Therefore, plant breeding has been investigated as a means to improve crop quality, by increasing crop concentrations of desirable trace elements and reducing those of potentially harmful trace elements such as cadmium (Cd) (Graham and Welch, 1999; Huang and Graham, 2001; Gregorio, 2002; Welch and Graham, 2004; Graham et al., 2007). Cultivar selection is an attractive method for changing the trace element profile of crops, as the benefit will persist in the seed and can reduce the requirement for other management techniques such as fertilization management or crop rotations.

Cadmium (Cd) is a trace element that is present in the soil naturally and from anthropogenic sources, including atmospheric deposition, application of sewage sludges and manures, irrigation water, and in fertilizers and soil amendments (Alloway and Steinnes, 1999). Cadmium can accumulate in the human body over time from ingestion of food containing Cd, leading to a risk of chronic toxicity with excessive intake. It is desirable to limit the concentration of Cd in crops used for human consumption in order to reduce potential health risks. Therefore, plant breeding has been investigated as a method of reducing Cd accumulation in a variety of crop species.

### 1.1. Differences among species

Crop species and cultivars differ widely in their ability to absorb, accumulate and tolerate Cd. (Bingham et al., 1975; Kuboi et al., 1986; Li et al., 1995b, 1997; Grant et al., 1999, 2000; Hocking and McLaughlin, 2000; Clarke et al., 2002; Clemins et al., 2002; Miller et al., 2006). Based on the concentration of Cd in commercially grown crops destined for international trade, crops such as sunflower, flax, rice and durum wheat have been identified as accumulators of Cd, frequently containing more than 0.10 mg Cd kg<sup>-1</sup> dry matter (Erdman and Moul, 1982; Li et al., 1997; Clarke et al., 1997a,b; Hocking and McLaughlin, 2000; Arao and Ishikawa 2006). The concentration of Cd in peanut, potato and soybean may also be of concern, particularly where they comprise a large portion of the diet (McLaughlin et al., 1994b; Bell et al., 1997; Arao and Ishikawa, 2006). In the case of peanut, the seed coat (testa) contains more than 10 times higher Cd concentration than the seed;

blanched seed without testa can meet crop Cd limitations more easily than whole roasted peanuts (Bell et al., 1997; McLaughlin et al., 2000). Spring wheat, barley, oat and maize generally contain Cd concentrations below 0.10 mg kg<sup>-1</sup> (Erdman and Moul, 1982; Hinesley et al., 1978; Oliver et al., 1995; McLaughlin et al., 2000).

The amount of Cd that enters the human diet from a crop depends on the amount of Cd accumulated in the portion of the plant that is consumed rather than solely on total plant uptake. Distribution of Cd within the plant is influenced by transport from roots to the shoots via the xylem, transfer from the xylem to the phloem and transport through the phloem from sources to sinks (Riesen and Feller, 2005). Both accumulation and distribution of Cd in the plant differs depending on the species, cultivar and growing conditions (Williams and David, 1973; Jackson and Alloway, 1992; Florijn et al., 1992; Florijn and Van Beusichem, 1993; Buckley et al., 1997; Grant et al., 1998, 1999; Harris and Taylor, 2001; Arao et al., 2003; Harris and Taylor, 2004; Arao and Ishikawa, 2006) and by the presences of other elements (Herren and Feller, 1997). Stolt et al. (2003) suggested that the larger accumulation of Cd in the grain of durum wheat as compared to bread wheat was associated with a higher total uptake by the plant. Differences among plant cultivars in secretion of low molecular weight organic acids may influence root uptake of Cd (Liu et al., 2007b). In other crops, Cd may be readily taken up, but restricted translocation from root to stem and to leaves and fruit may result in proportionally lower concentrations in the fruits, storage roots/tubers and grains than in the root or leaves (Florijn and Van Beusichem, 1993; Guo and Marschner, 1996; Buckley et al., 1997; Hart et al., 1998; Archambault et al., 2001; Harris and Taylor, 2001, 2004; Arao and Ishikawa, 2006). Accumulation of large amounts of Cd in the root may limit the accumulation of Cd in edible above-ground portions of the plant (Buckley et al., 1997; Sugiyama et al., 2007). In studies where shoots and rootstocks from high and low accumulating soybean lines were recombined by grafting, rootstocks with a capacity to accumulate high amounts of Cd led to a reduction in the Cd concentration of seed, indicating that the accumulation of Cd in the seed was reduced by high accumulation in the root and was controlled by the rootstock cultivar (Sugiyama et al., 2007). Transport of Cd within the plant appears to be largely phloem-mediated (Reid et al., 2003; Dunbar et al., 2003; Tanaka et al., 2007), although the relative phloem-mobility of Cd may be less than that of Zn (Riesen and Feller, 2005). Cadmium was measured in the phloem sap of rice, collected using cut stylets of the brown planthopper, at concentrations that increased with Cd application in the nutrient solution (Tanaka et al., 2003), providing direct evidence for translocation of Cd via the phloem. In subsequent studies measurement of distribution ratios of <sup>109</sup>Cd between rice grain and glumes indicated that more than 90% of the Cd in the grains had been translocated via the phloem (Tanaka et al., 2007), although the very high-Cd concentrations used in the nutrient solutions may have influenced the Cd distribution. Differences among species in Cd concentration of the seeds may be in part related to differences in the abilities of plants to control movement of Cd from the xylem into the phloem and via the phloem to the seeds (Hocking and McLaughlin, 2000; Cakmak et al., 2000a,b; Hart et al., 2002, 2006; Tanaka et al., 2007).

## 1.2. Differences among cultivars

Wide variations in the concentration of Cd among cultivars have been documented in many species, including, but not restricted to asparagus bean (*Vigna unguiculata* subsp. *Sesquipedalis* L.) (Zhu et al., 2007), barley (Chang et al., 1982; Chen et al., 2007), carrot (Harrison, 1986), corn (maize) (Hinesly et al., 1978), cucumber (Harrison and Staub, 1986), common wheat (Kjellström et al., 1975; Andersson and Pettersson, 1981; Gray et al., 2001; Stolt et al., 2006), durum wheat (Oliver et al., 1995; Clarke et al., 1997a,b, 2002; Li et al., 1997; Greger and Löfstedt, 2004; Stolt et al., 2006), linseed (flax) (Hocking and McLaughlin, 2000; Grant et al., 2000), lettuce (Crews and Davies, 1985; Thomas and Harrison, 1991), oat (Tanhuanpää et al., 2007), pea (Rivera-Becerril et al., 2002), peanut (Bell et al., 1997; McLaughlin et al., 2000), potato (McLaughlin et al., 1994b; Dunbar et al., 2003), rice (Arao and Ae, 2001, 2003; Arao and Ishikawa, 2006), ryegrass (Gray and McLaren, 2005), soybean (Arao et al., 2003; Ishikawa et al., 2005a; Arao and Ishikawa, 2006), and willow (Granel et al., 2002).

From a health perspective, the major concern for Cd accumulation occurs with staple crops that may form a large proportion of the human diet, and hence a large proportion of the dietary Cd exposure, such as rice, wheat, potato and possibly soybean. As a component of pasta and couscous, durum wheat forms a large part of many diets. Analysis of durum wheat cultivars grown in a variety test in South Dakota showed that grain Cd concentration ranged from 0.13 to 0.25 mg kg<sup>-1</sup> with an average of 0.22 mg kg<sup>-1</sup> (Erdman and Moul, 1982). Analysis of wheat cultivars grown in Interstate Wheat Variety trials across Australia between 1988 and 1989 and in Western Australia in 1990–1992 showed significant genetic difference among cultivars in grain Cd concentration (Oliver et al., 1995). Low-Cd cultivars tended to have similar pedigrees, indicating potential for selecting lines for low-Cd concentration. Cadmium concentration varied widely in the grain of bread wheat and durum wheat cultivars grown in variety evaluation trials on uncontaminated soils on the Canadian prairies (Clarke, unpublished). Grain Cd concentrations were measured in Arcola and Kyle durum wheat and Genesis and Katepwa spring wheat (*T. aestivum* L.) from 12 trials in Saskatchewan. Cadmium concentration averaged 0.077 mg kg<sup>-1</sup> for Arcola, 0.157 for Kyle, 0.038 for Genesis and 0.055 for Katepwa. While there was a three-fold range in Cd concentrations with location, the cultivar ranking was maintained across locations. Differences in Cd in the grain among durum wheat cultivars appeared to be associated with greater retention of Cd in the root in low-Cd cultivars (Stolt et al., 2003; Chan and Hale, 2004; Harris and Taylor, 2004; Hart et al., 2005, 2006). In solution culture studies, Greger and Löfstedt (2004) observed that cultivars of durum wheat differed in Cd concentration attributable to differences in translocation from the root to the shoot and within the shoot, rather than to differences in root uptake.

Potatoes are another crop that can form a significant portion of the diet. McLaughlin et al. (1994b) reported that mean tuber Cd concentrations differed significantly in 14 commercial potato cultivars, and suggested that tuber Cd concentrations could be reduced by up to 50% by appropriate choice of cultivar. As with durum wheat, differences in Cd

concentration in potato tubers between cultivars were associated with differential distribution of Cd within the plant rather than with differences in the total plant accumulation of Cd (Table 1) (Dunbar et al., 2003).

Cadmium concentration has also been shown to vary among rice cultivars (Arao and Ae, 2001, 2003; Ishikawa et al., 2005a; Arao and Ishikawa, 2006). Arao and Ae (2003) evaluated 49 cultivars of rice grown in containers under simulated upland conditions on soils contaminated with Cd. Differences among cultivars were large and consistent between the soils. In addition, relative rankings of five cultivars was similar when they were evaluated in pots simulating either upland or paddy conditions. Differences among rice cultivars in Cd concentration in the grain were much higher than cultivar differences in Cd concentration in the roots, stems and leaves (Liu et al., 2007a). Genetic differences occurred in the translocation of Cd from the shoot to the grain in rice (Arao and Ishikawa, 2006). He et al. (2006) also reported substantial genetic variation in the Cd concentration of lowland rice (0.06 to 0.99 mg Cd kg<sup>-1</sup>) grown on contaminated soils but indicated that the differences appeared to be driven by Cd uptake by the plant rather than by differential partitioning of the Cd among plant parts.

There was also considerable genetic variation observed among soybean cultivars (Bogges et al., 1978; Bell et al., 1997; Arao et al., 2003; Arao and Ishikawa, 2006), with low-Cd cultivars appearing to retain more Cd in the root and translocate less to the seed than high-Cd cultivars (Ishikawa et al., 2005a).

Sunflower and flaxseed (linseed) form a much lower portion of the human diet than do rice, wheat, potatoes or soybean. However, sunflower and flaxseed tend to accumulate relatively high concentrations of seed Cd and there is concern that trade in these two crops may be restricted due to excess Cd concentration. Considerable genetic variation occurred among sunflower lines grown across a range of soils (Li et al., 1995b, 1997; Miller et al., 2006). Li et al. (1995b) reported large genetic differences in kernel Cd concentration among 200 sunflower genotypes grown at four locations. On average, Cd

**Table 1 – Comparison of growth, total plant Cd and concentrations of Cd in tubers and whole plant tissue of potato cvs**

	Wilwash	Kennebec	Significant difference
Total dry weight (g)	156±15	159±10	Ns
Total plant Cd (mg)	47.6±4.3	48.9±3.1	Ns
Plant Cd concentration (mg g DW)	0.308±0.024	0.282±0.003	Ns
Tuber yield (g DW plant)	143±14	173±10	Ns
Tuber Cd concentration (mg g DW)	0.143±0.13	0.236±0.009	P<0.001
Kennebec and Wilwash (Dunbar et al., 2003). Values are the mean and SE of 4 plants harvested 13 weeks after planting.			



concentration in the five genotypes with lowest Cd concentration was four-fold lower than that in the five genotypes with the highest Cd concentration. Linseed also shows relatively large genetic variation in Cd concentration (Marquard et al., 1990; Cieslinski et al., 1996). A field screening of more than 2700 accessions from the USDA flax collection on a Fargo silty clay soil known to be relatively high in plant available Cd showed a range in seed Cd of 0.27 to 3.60 mg kg<sup>-1</sup> (Hammond et al., 1999). In Australian studies, linseed lines showed a 2.3-fold variation in Cd concentration (Hocking and McLaughlin, 2000). Variation also occurred within flax (linseed) genotypes grown in Manitoba, Canada, with seed Cd concentrations ranging from 0.87 (cv. FP 948) to 1.96 (cv. Vimy) mg kg<sup>-1</sup> (Kenaschuk and Dribnenki, pers. comm., Morden, MB, Canada).

### 1.3. Breeding and selection of low cadmium crops

The fact that genetic variability exists with a species in the tendency to accumulate Cd provides an opportunity to utilize plant breeding to select for genetically low-Cd concentration. In the absence of selection for the low-Cd trait new cultivars may be randomly either higher or lower than traditional cultivars. For example, when modern sunflower hybrids were developed, it appears that the genotypes converted to inbreds had higher kernel Cd concentrations than random sunflower genotypes (Li et al., 1995b). Similarly in flax, the Cd content of new cultivars may be higher or lower than those in traditional lines depending on the exact genetic history of the new strain (Marquard et al., 1990).

Cultivar selection is an important method to limit Cd uptake and accumulation in crops, but the process is long and complex. Initially breeders must: 1) find genetic variation in the Cd concentration in existing germplasm; 2) learn the inheritance of the low-Cd genetic character; 3) develop a breeding strategy to combine low-Cd traits with high yields, disease resistance and other quality traits in modern cultivars; and 4) develop inexpensive methods to combine the low-Cd characteristic with other desired traits. Identifying low-Cd phenotypes by analysis of the grain is more costly than many other conventional breeding activities, due to the high cost of analysis.

Although genotypic differences in Cd concentrations have been studied in a range of crops, only limited effort has been made in the past to utilize plant selection or breeding to reduce the Cd in crops. Recently, because of market forces, researchers have placed greater emphasis on producing low-Cd cultivars of several grain crops, including durum wheat (Penner et al., 1995; Li et al., 1997; Clarke et al., 1997a, 1997b, 2002, 2005, 2006; Archambault et al., 2001), sunflower (Li et al., 1995b,c, 1997; Miller et al., 2006), rice (Arao and Ae, 2001, 2003; Liu et al., 2003, 2005, 2006, 2007a; Ishikawa et al., 2005a; Arao and Ishikawa, 2006; He et al., 2006; Yu et al., 2006) and soybean (Arao and Ae, 2001; Arao et al., 2003; Ishikawa et al., 2005a; Arao and Ishikawa, 2006).

In Canada, a breeding program was established in 1991 to reduce the Cd content of Canadian durum wheat. Initial surveys in the early 1990s for variation in the Cd concentration in domestic Canadian cultivars and lines showed that the cultivar Hercules and its derivative Arcola had a lower grain Cd

concentration than other Canadian cultivars, with the concentration in Arcola being approximately half that in the predominant Canadian durum cultivar Kyle. Subsequent surveys of durum germplasm identified considerable genetic variation in grain Cd concentration of cultivars and lines introduced from sources such as CIMMYT and ICARDA international nurseries.

An understanding of the inheritance and heritability of the genetic variability was important in designing the breeding program. Grain Cd concentration was found to be controlled by a single gene, with low-Cd dominant in the crosses studied (Clarke et al., 1997b). This was later shown to reside on chromosome 5B (Knox et al., 2003). Lines with the low-Cd trait had restricted root to shoot translocation, which limited the Cd accumulation in the grain (Harris and Taylor, 2001; Chan and Hale, 2004). The reduced translocation was associated with lower Cd concentration in the xylem sap and reduced xylem sap exudation (Fig. 1) (Harris and Taylor, 2004). The low-Cd trait was highly heritable so selection on a single plant basis was feasible. Cadmium concentration of leaf tissue was highly correlated with grain Cd concentration ( $r=0.87$  to  $0.89$ ) and therefore could predict the plant phenotype, which would be useful in backcrossing the low-Cd trait into high-Cd cultivars (Clarke et al., 1997b).

The crossing program developed near-isogenic high/low grain Cd concentration lines from five durum wheat crosses

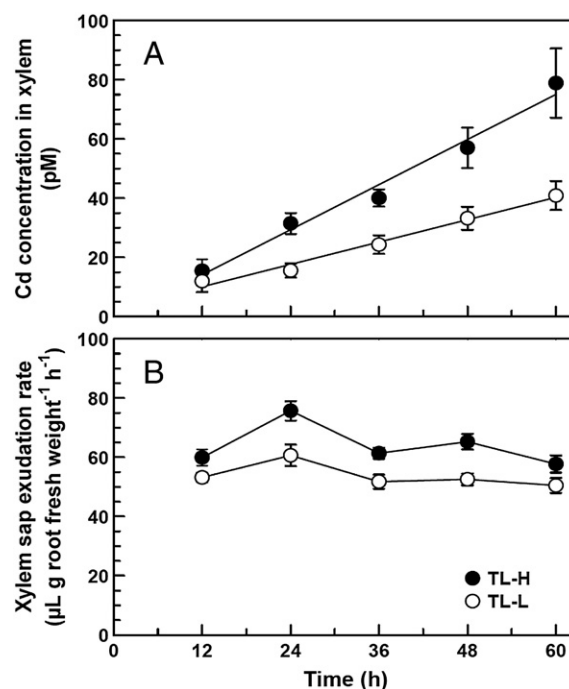


Fig. 1 – <sup>109</sup>Cd transport in xylem sap of durum wheat seedlings. Time-course of <sup>109</sup>Cd concentration in xylem sap (A) and the rate of xylem sap exudation (B) of high (TL-H) and low (TL-L) Cd-accumulating isolines of durum wheat. Roots of 6-d old seedlings were exposed for between 12 and 60 h in 15 mL of complete nutrient solution containing 25 pM <sup>109</sup>Cd (solutions changed every 12 h). Shoots were then excised 5–7 mm above the roots and xylem sap collected for 6 h. Means and standard errors of 10 replicates are plotted (Harris and Taylor, 2004).

**Table 2 – Grain Cd concentration (mg kg<sup>-1</sup>) of durum isogenic pairs and parents grown in varying environments between 1994 and 1996 (Adapted from Clarke et al., 2002)**

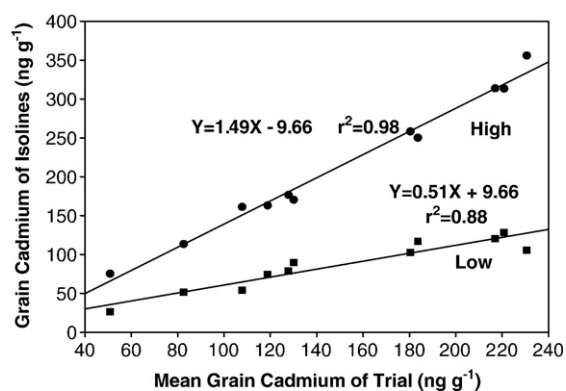
Genotype <sup>a</sup>	1994		1995			1996			
	Swift Current	Fargo	Regina	Swift Current	Stewart Valley	Casselman	Regina	Swift Current	Stewart Valley
8982-SF-L	0.08	0.09	0.11	0.12	0.06	0.09	0.11	0.05	0.06
8982-SF-H	0.24	0.36	0.16	0.27	0.19	0.21	0.33	0.11	0.15
8982-TL-L	0.09	0.08	0.07	0.12	0.08	0.10	0.11	0.04	0.08
8982-TL-H	0.24	0.27	0.19	0.35	0.13	0.28	0.34	0.11	0.18
W9260-BC-L	0.19	0.15	0.15	0.18	0.11	0.14	0.19	0.10	0.03
W9260-BC-H	0.29	0.41	0.19	0.34	0.19	0.30	0.32	0.14	0.07
W9261-BG-L	0.11	0.12	0.05	0.10	0.06	0.10	0.14	0.05	0.13
W9261-BG-H	0.24	0.41	0.17	0.27	0.15	0.27	0.27	0.10	0.23
W9262-339A-L	–	0.09	0.07	0.09	0.07	0.08	0.10	0.03	0.20
W9262-339A-H	–	0.34	0.14	0.33	0.16	0.23	0.30	0.11	0.07
Biodur	–	0.08	0.04	0.07	0.05	0.09	0.05	0.03	0.03
DT471	0.12	0.13	0.06	0.13	0.77	0.11	0.11	0.06	0.07
DT618	0.31	0.37	0.25	0.33	0.20	0.29	0.37	0.13	0.23
DT630	0.20	0.38	0.11	0.25	0.15	0.32	0.27	0.10	0.13
Kyle	0.24	0.33	0.19	0.30	0.17	0.23	0.26	0.10	0.20
Nile	0.09	0.08	0.09	0.09	0.06	0.12	0.13	0.04	0.07
Mean	0.19	0.23	0.13	0.21	0.16	0.18	0.21	0.08	0.12
SED <sup>b</sup>	0.03	0.02	0.04	0.04	0.02	0.02	0.02	0.01	0.02
Contrast H vs. L <sup>c</sup>	**	**	**	**	**	**	**	**	**

<sup>a</sup> Isogenic line designations ending 'L' indicate low-Cd accumulator, and 'H' indicates high accumulator.

<sup>b</sup> Standard error of a difference.

<sup>c</sup> Significance of single degree of freedom contrast of all high vs. all low isolines: \*P<0.05, \*\*P<0.01.

(Clarke et al., 1997a). Each high/low pair was genetically uniform except for the Cd concentration trait. In studies in Saskatchewan and North Dakota from 1994–96, the average grain Cd concentration was approximately 2.5 times greater for the high than for the low isolines (Table 2) (Clarke et al., 2002). This difference varied with soil and climatic conditions, being smaller in environments which generally produced a low grain Cd concentration, and greater in environments that produced a high-Cd concentration (Fig. 2). The low-Cd uptake trait had no significant effect on average yield, grain protein concentration, test weight, kernel weight, days to maturity or lodging score (Clarke et al., 2002) as indicated by comparison of the high- and low-



**Fig. 2 – Grain cadmium concentration of high and low cadmium near-isogenic lines grown at five locations in Saskatchewan and North Dakota in 1994 to 1996 (Clarke et al., 2002).**

Cd isolines. The low-Cd uptake trait had no consistent effect on grain concentration of other elements such as zinc, wherein the high and low lines did not differ in seven of eight environments. However, there was some indication that the low-Cd trait may also be associated with a reduction in Zn accumulation in grain under Zn-deficient conditions in a solution culture experiment (Hart et al., 2005). The low-Cd characteristic has been incorporated into newly registered durum wheat cultivars (Clarke et al., 2005, 2006). The first commercially-successful low-Cd cultivar, Strongfield (Clarke et al., 2005), was released in 2004 and is now sown on more than 25% of the durum area in Canada. All new durum cultivars released in Canada must carry the low-Cd trait.

A similar program was undertaken to reduce the Cd concentration in sunflowers after a guide value (Richtwert) of

**Table 3 – Seed Cd concentration in standard and low-Cd maintainer and restorer lines and hybrids of confectionary sunflower (adapted from USDA-ARS, 2003)**

	Maintainer lines		Restorer lines		Hybrids	
	Line	Seed Cd (mg kg <sup>-1</sup> )	Line	Seed Cd (mg kg <sup>-1</sup> )	Hybrid	Seed Cd (mg kg <sup>-1</sup> )
Standard selections	HA 285	1.34	RHA 294	1.19	Hybrid 924	1.38
	HA 323	0.88	RHA 324	0.70		
	HA 448	0.75	RHA 450	0.55	HA448/RHA 450	0.68
Low-Cd selections	HA 449	0.64			HA449/RHA 450	0.67

0.6 mg Cd kg<sup>-1</sup> dry weight (DW) for Cd in sunflower used for confectionary purposes was established by Germany. Confectionary sunflower seed had values reaching 1.33 mg kg<sup>-1</sup> DW when grown in North Dakota and Minnesota on poorly drained, fine textured soils, containing high background concentrations of chloride (Li et al., 1995a). Two hundred genotypes, including USDA-ARS germplasm lines, plant introductions from various countries, and interspecific germplasm lines were grown in 1994 in replicated field trials at four locations (Li et al., 1995b,c). Several accessions were identified that contained very low-Cd and a maintainer and a restorer line among the USDA-ARS confection germplasm lines were found to be intermediate in Cd uptake. The breeding program initiated in North Dakota has resulted in the release of lines with Cd concentrations substantially below the average value for Cd in sunflower (Table 3) (USDA-ARS, 2003; Miller et al., 2006). Use of these lines as parents produced hybrids with a 50% decrease in kernel Cd concentration.

Reducing the Cd concentration of rice is of particular importance because a major health problem has been as-

sociated with consuming rice with high concentrations of Cd (Chaney et al., 1999, 2004). Considerable variability in the Cd concentration has been found in rice (Table 4) including Japonica, Indica and hybrid rice (Morishita et al., 1987; Arao and Ae, 2001, 2003; Liu et al., 2003, 2005, 2006; Cui et al., 2004; Ishikawa et al., 2005a; Arao and Ishikawa, 2006; He et al., 2006; Yu et al., 2006) and breeding programs have been initiated to select for low-Cd cultivars. The relative ranking of rice cultivars appears to be similar under paddy and upland conditions (Arao and Ae, 2003), thus the low-Cd trait should persist when the cultivars are grown in different environments. Additionally, high-Cd lines of Japonica-Indica hybrid rice are being investigated for their potential in phytoremediation of contaminated sites (Ae and Arao, 2002). Recently, Ishikawa et al. (2005b) reported identification of the putative quantitative trait loci for Cd concentration in brown rice, which should facilitate the breeding of new cultivars of low-Cd rice.

Genetic variability in soybean (Guo and Marschner, 1996; Bell et al., 1997; Arao and Ae, 2001; Arao et al., 2003; Ishikawa

**Table 4 – Mean grain Cd concentration of rice cultivars grown in two soils (container experiments, 2000) (Arao and Ishikawa, 2006)**

Cultivar name	Country of origin	Soil A (mg kg <sup>-1</sup> dw)				Soil B (mg kg <sup>-1</sup> dw)			
LAC 23	SLE	0.19	a			1.06	a		
HU-LO-TAO	CHN	0.39	a	b		0.79	a		
AKITAKOMACHI	JPN	0.66	a	b	c	1.28	a	b	
NIPPONBARE	JPN	0.71	a	b	c	1.71	a	b	c
SASANISHIKI	JPN	0.80	a	b	c	1.16	a	b	
KOSHIHIKARI	JPN	0.80	a	b	c	1.83	a	b	c
BATATAIS	BRZ	0.87	a	b	c	1.46	a	b	c
KETAN	IDN	0.88	a	b	c	2.36	a	b	c
AKIHIKARI	JPN	0.96	a	b	c	1.05	a		
CAIAPO	BRZ	1.02	a	b	c	1.88	a	b	c
PEROLA	BRZ	1.03	a	b	c	1.58	a	b	c
GURANI	unknown	1.06	a	b	c	1.69	a	b	c
DHILI BORO	BGD	1.22	a	b	c	3.28	a	b	c
DENNYUIGOU	JPN	1.24	a	b	c	2.78	a	b	c
KHAO DAM	LAO	1.36	a	b	c	2.73	a	b	c
MIZUHATAMOCHI	JPN	1.48	a	b	c	3.94	a	b	c
BAARAN BORO	BGD	1.54	a	b	c	2.29	a	b	c
GHARIB	IRN	1.56	a	b	c	2.58	a	b	c
IRAT212	CIV	1.62		b	c	2.71	a	b	c
SHORT GRAIN	THA	1.67		b	c	7.24			f
BG1	JPN	1.78		c	d	2.50	a	b	c
TSUKUBAHATAMOCHI	JPN	1.87		c	d	2.14	a	b	c
NOURIN24GOU	JPN	1.89		c	d	2.76	a	b	c
GERDEH	IRN	1.90		c	d	2.23	a	b	c
KUROMOKU	JPN	2.15		d	e	2.80	a	b	c
KASALATH	IND	2.22		e	f	2.81	a	b	c
HATAKINUMUCHI	JPN	2.34		f	g	2.27	a	b	c
CHIYOMMINORI	JPN	2.36		f	g	4.85		c	d
CHAHORA 144	PAK	2.42		g	h	4.51	b	c	d
RD7	THA	2.58		h	i	6.16		d	e
LMN111	THA	2.66		i	j	7.05			f
PEH-KUH-TSAO-TU	TWN	2.98		j	k	6.66		e	f
IR-8	PHL	3.27		k	l	7.65			g
HABATAKI	JPN	3.56		l	m	6.94			f
MILYANG23	KOR	4.31			m	7.18			f
Average		1.70				3.25			

Means followed by the same letter are not significantly different at the 5% level according to the Tukey HSD test.

BGD: Bangladesh, BRZ: Brazil, CHN: China, CIV: Cote d'Ivoire, IDN: Indonesia, IND: India, IRN: Iran, JPN: Japan, KOR: Korea, LAO: Laos, PAK: Pakistan, PHL: Philippines, SLE: Sierra Leone, THA: Thailand, TWN: Taiwan.

et al., 2005a; Arao and Ishikawa, 2006) and flax (Cieslinski et al., 1996; Li et al., 1997; Hocking and McLaughlin, 2000; Grant et al., 2000) has been reported. Cadmium concentration in young tissue of soybean was correlated well to the final Cd concentration of the mature seed, which would facilitate breeding (Arao and Ishikawa, 2006). Based on the importance of soybean as a staple food crop, development of low-Cd soybean cultivars should be a priority (Arao et al., 2003; Ishikawa et al., 2005a; Morrison, 2005; Arao and Ishikawa, 2006). In contrast, only small amounts of flaxseed are normally consumed in the human diet. While there is interest in developing low-Cd flaxseed to encourage its consumption as a health-promoting food, a breeding program for reduced Cd is considered to be of lesser importance than in the staple crops such as durum wheat, rice and soybean.

#### 1.4. Challenges to use of genetic selection for reducing Cd concentration in crops

While cultivar selection can be effective in reducing the potential Cd concentration in crops, there are still constraints in utilizing this approach to produce low-Cd cultivars. Development and testing of a new cultivar is time-consuming, because the low-Cd characteristic must be incorporated into a cultivar that has acceptable characteristics for yield, agronomic suitability, quality and disease resistance. For example, it was 10 years from the initiation of the Canadian breeding program for low-Cd durum wheat to the release in 2001 of the first low-Cd cultivar, named AC Napoleon. It is estimated that it will take 5 to 10 years to develop a new rice cultivar acceptable to Japanese consumers by crossing low-Cd alien rice varieties with acceptable Japonica ones (Ae and Arao, 2002). The process of selecting for low-Cd may be more difficult and time-consuming in out-crossed crops than in crops that are self-pollinated.

Chemical analysis for Cd is expensive and is required to confirm that the lines are actually low in Cd. An ability to detect and select for genetic differences in Cd concentration at an early growth stage will reduce the time and cost of the breeding program (Arao and Ae, 2001; Archambault et al., 2001; Arao and Ishikawa, 2006; Stolt et al., 2006). A RAPD marker developed for use in durum wheat (Penner et al., 1995) improved the efficiency of the screening process and is considerably less expensive than chemical analysis. The marker, which is in repulsion with the low cadmium allele, can be used with appropriately-configured crosses. Markers have also been found associated with a major gene affecting Cd accumulation in oat (Tanhuanpää et al., 2007). The cost of selecting cultivars in a breeding program declines as more breeding lines carry the low-Cd trait. For example, the Canadian durum programs now require much less Cd testing than when the program was initiated because a large pool of low-Cd germplasm is available for use in new crosses. However, the initial phase of conversion to low-Cd was costly in terms of Cd analyses as well as in the fact that less progress in breeding for other economic traits due to the inclusion of low-Cd as an essential selection characteristic.

Inclusion of low-Cd as a selection criterion adds an additional factor to an already lengthy list of characteristics that have to be present in a potential new cultivar. As well as the basic characteristics of yield, quality and disease resis-

tance, factors such as herbicide tolerance, lodging resistance, drought resistance, insect resistance, and days to maturity are considered. There is also pressure to select for characteristics such as nutrient use efficiency and nutritional quality. The value of investing time and resources in selection for the low-Cd characteristic must be assessed relative to investment in selection for other characteristics when determining breeding priorities. In addition, Cd concentration in crops may be affected by other selection activities. For example, in selecting for aluminum (Al) tolerance in crops growing on acid soils (pH < 5.5), it may be necessary to incorporate genes to limit the large Cd uptake that would occur at that pH. Also, when screening to improve the concentration of bioavailable zinc (Zn) in grains, it is important to verify that Cd is not also increased because of the similarity of these elements in some biological processes.

While selection can produce low-Cd cultivars, the Cd concentration of both low- and high-Cd cultivars will be influenced by both soil and management practices (Grant et al., 1999). When low- and high-Cd isolines of durum wheat were grown on a variety of soils and environments, the low-Cd lines were consistently lower than the high-Cd lines at each site-year, but both low- and high-Cd cultivars produced Cd concentrations near the proposed  $0.2 \text{ mg kg}^{-1}$  limit at some locations (Table 2). Similarly, although the Cd concentration of McGregor flaxseed grown in Manitoba, Canada was lower than that of Vimy, both cultivars contained excessive Cd concentrations when grown at a location where there was a large amount of phytoavailable Cd (Grant et al., 2000). Application of fertilizers, addition of Cd in fertilizers, biosolids or other soil amendments, the use of high-chloride irrigation water, soil salinity and soil acidification can all increase Cd phytoavailability in soil (He and Singh, 1994; McLaughlin et al., 1994a, 1995; Smolders and McLaughlin, 1996, 1998; Grant and Bailey, 1997; Grant et al., 1999). Correction of Zn deficiencies, flooding of rice paddies combined with application of organic matter and possibly liming or addition of organic residues can reduce Cd uptake by crops (Abdel-Sabour et al., 1988; Xue and Harrison, 1991; Choudhary et al., 1994; Oliver et al., 1994; McLaughlin et al., 1995; Grant and Bailey, 1997; Sparrow and Salardini, 1997; Grant et al., 1999, 2000; Welch et al., 1999; Cakmak et al., 2000a; Hart et al., 2002, 2005; Jiao et al., 2004; Kölel et al., 2004). Combining management practices that limit Cd accumulation with use of low-Cd cultivars would be more effective at reducing Cd movement into the food chain than growing low-Cd cultivars alone. Although management practices and use of appropriate cultivars can decrease Cd in crops there is still a risk of long-term accumulation of phytoavailable Cd in agricultural soils that could increase the Cd concentration in both low- and high-Cd cultivars.

The risk of toxicity from Cd in food is influenced not only by Cd concentration but also by concentrations of other trace elements such as Zn and iron (Fe) (Chaney et al., 1999). Breeding programs are underway to increase the concentration of essential trace elements to enhance the nutritional value of staple crops (Graham and Welch, 1999; Huang and Graham, 2001; Welch and Graham, 2002, 2004; Graham et al., 2007). Breeding programmes to increase concentrations of essential trace elements would have the combined benefit of enhancing the nutritional value of staple crops while reducing



the bioavailability of Cd, particularly if low-Cd was included as a selection criterion.

## 2. Conclusions

Plant breeding programs have utilized natural genetic variation within crop species to select durum wheat cultivars and sunflower hybrids used for confectionary purposes that accumulate less than average Cd concentrations. Breeding programs are underway to produce low-Cd cultivars of rice and soybean. The potential also exists to produce low-Cd cultivars of many other food crops if this is perceived as a priority. In durum wheat, low-Cd concentration is controlled by a single dominant gene. The allele for low-Cd concentration does not appear to affect major economic traits and should not cause problems when incorporated into durum cultivars. Selection of increased concentrations of essential trace elements such as Fe and Zn may also reduce the bioavailability of Cd, while enhancing the nutritional value of crops. The Cd concentration in both low- and high-Cd cultivars can increase if environmental factors, soil salinity, high-Cl<sup>-</sup> irrigation water, or management practices increase phytoavailable Cd. While growing low-Cd cultivars of food crops can reduce the risk of movement of Cd into the human diet, accumulation of Cd in soils may still be a concern for the long-term sustainability of crop production and quality.

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